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TITLE: Multidrug Resistance in Breast Cancer: Occurrence and Therapeutic Implications

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13. ABSTRACT (Maximum 200) Although clinical development of drug resistance during treatment of breast cancer has been apparent for at least two decades, little known about the mechanisms involved. Multidrug resistance (mdr) mediated by the MDR1 gene expression is one of multiple factors identified which may be associated with declining therapeutic efficacy of anticancer drugs after failure of an initial therapy. This research program seeks to define MDR1 expression in clinical specimens, and its relationship to age, menopausal status, stage, hormone receptors, site of disease, and prior treatment. Through clearly defined laboratory and clinical protocols, it will also correlate drug treatment of breast cancer to the expression and function of MDR1 and its P-glycoprotein (Pgp). Clinical specimens from primary or metastatic breast cancer patients are tested for MDR1 expression by Polymerase Chain Reaction (PCR) and by immunostaining for Pgp with monoclonal antibodies. These analyses are also applied to patients enrolled in a series of therapeutic studies including paclitaxel (Taxol TM) in patients having failed other therapies. Preliminary expression analysis of other potentially important genes, such as p53 tumor suppressor gene and p7 is in progress. Response to treatment and laboratory findings are correlated. In patients exhibiting resistance to Taxol, strategies involving resistance-reversal through dose-scheduling and resistance-modifiers are being developed. A randomized Phase II trial of Taxol without and with resistance modifiers is in progress. The composite of these studies will provide information defining the importance of multidrug resistance in determining the outcome from systematic taxol therapy in the treatment of breast cancer.			
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FOREWORD

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INTRODUCTION.

The systemic treatment of breast cancer has been hampered by the eventual emergence of drug-resistant populations, in spite of initial responses in the majority of patients. In addition to anthracyclines, alkylating agents, flouropyrimidines and other antimetabolites, in the past 5 years the taxanes have emerged as drugs with major antitumor activity both in the absence and the presence of previous treatment. Since paclitaxel is an excellent substrate of the Pgp-efflux pump encoded by the multidrug resistance gene MDR1 and its role in breast cancer treatment is undergoing extensive evaluation, trials with this drug represented an excellent opportunity to delineate the importance of this drug-resistance mechanism in this disease. Therefore, the grant was structured to focus on determinations of Pgp before and after treatment with anthracycline-containing regimens, and on clinical trials of paclitaxel with drugs known to reverse Pgp action and restore sensitivity. These MDR1-reversal drugs have included calcium-channel blockers, steroid hormone-related derivatives, cyclosporins, and a number of other drug classes. The four tasks of the grant included 1) delineating the distribution of Pgp in various subsets of patients with breast cancer (premenopausal, postmenopausal, stage related, hormone receptor related), 2) developing a paclitaxel plus a resistance reversal drug regimen in the treatment of advanced breast cancer, 3) performing a randomized clinical trial to assess the strategy of adding a multidrug resistance reversal to paclitaxel, and 4) investigating new methods to identify and reverse multidrug resistance.

The identification of the circumstances surrounding the occurrence of multidrug resistance in breast cancer has become even more important since the grant funding began in October 1994. Two of the drug classes that are key components in the current systematic treatment of breast cancer, the taxanes and the anthracyclines have been the focus of much clinical work, both in the adjuvant and in the advanced disease setting. Cooperative group trials are investigating adjuvant doxurubicin as sequentially versus combined with cyclophosphamide in high risk patients with 0 to 3 lymph nodes involved (SWOG-9313), or in arms with escalating doses, with or without paclitaxel for node positive disease (SWOG-9410, intergroup study). In advanced breast cancer, combinations of doxorubicin and paclitaxel are being compared to either drug alone (Intergroup study, coordinator Dr. George Sledge). While awaiting the results of this study, pilot studies have indicated not only striking activity, but also the likelihood of pharmacokinetic interactions between the two drugs with the potential of leading to higher AUCs of both doxorubicin and its metabolite, doxorubicinol. Presumably, in part related to this interaction, a propensity to enhanced cardiotoxicity has been described (1-2). The pharmacokinetic interactions are likely to occur because both doxorubicin and paclitaxel are excellent substrates for MDR1-P-glycoprotein (Pgp). In the development of future therapeutic strategies it is highly desirable that the circumstances surrounding the expression of MDR1 in these cancers be well characterized.

During this grant year there have been some administrative changes in the conduct of the research effort. Dr. Franco Muggia, Principal Investigator during years 01 - 03, accepted a position at New York University (NYU) and suggested that the co-Investigator of the grant, Dr. Michael Press, become the Principal Investigator during the remaining grant period. Dr. Muggia's clinical investigations related to this grant would continue at NYU under a subcontract from the University of Southern California (USC) to complete the clinical investigations. Prior to leaving USC to become director of NYU Breast Cancer Research Program, Dr. Muggia held a series of meetings with participants in this research effort, including the current principal investigator, Michael Press, personnel involved in the laboratory studies (Dr. Xiaowei Yang), personnel involved in the randomized clinical study that was undergoing final changes prior to activation and submission to the U.S. Army Medical Research and Materiel Command's Human Use Office (Dr. Darcy Spicer, and Dr. Susan Groshen, biostatistician in the project). The purpose of these meetings was to review research progress and expedite an orderly transition in the administration of these on-going investigations.

EXPERIMENTAL METHODS AND PROCEDURES.

Our progress during year 03 is summarized below. The report covers work conducted in the laboratory component of the study to characterize expression of the multi-drug resistance gene (*MDR-1*) and other genes as potential predictors of responsiveness to taxol chemotherapy.

Laboratory Studies.

Multi-Drug Resistance Gene Expression. P-glycoprotein (Pgp)-associated multidrug resistance (MDR) is related to intrinsic and acquired cross resistance to anthracyclines, vinca alkaloids and other antineoplastic antibiotics. The expression of *MDR1* gene and its gene product, P-glycoprotein, was investigated in primary and advanced breast cancers to assess the role of P-glycoprotein in clinical decision-making. Expression was assessed by immunohistochemistry in 121 breast cancers, by reverse transcription-polymerase chain reaction (RT-PCR) in 36 of these breast cancers and by Northern Blot in six representative tumors. Fifty-three of the women had received no treatment prior to surgical resection or biopsy of their breast carcinoma. Forty-eight women had received doxorubicin treatment and seventeen women had received either radiation and 5-fluorouracil treatment or other chemotherapy prior to tumor biopsy. Thirty-one of the 37 women whose breast cancers were analyzed by RT-PCR had previously been treated with doxorubicin chemotherapy.

Our initial efforts to demonstrate P-glycoprotein with RT-PCR were unsuccessful because we were running the polymerase chain reaction for both *MDR1* mRNA and our positive control gene beta-2 microglobulin in the same reaction mix. This is appropriate for semi-quantitative PCR analyses and analysis of mRNAs which are expressed at moderate levels. However, for rare mRNAs this approach permits the control gene, expressed in moderate abundance, to compete effectively for nucleotides in the reaction mix and prevent identification of rare mRNA. Therefore, we modified our PCR protocol to run the control and *MDR1* RT-PCR reactions in separate tubes. This did permit us to identify an *MDR1* product. Forty-four percent of breast cancers tested by this protocol showed *MDR1* mRNA expression. However, immunohistochemistry in these and 78 additional breast cancers failed to demonstrate P-glycoprotein in the carcinoma cells of the breast tumors but did demonstrate P-glycoprotein in connective tissue stromal cells, considered to be monocytes (macrophages or activated lymphocytes) by morphology. Immunohistochemistry did demonstrate physiological levels of P-glycoprotein expression in normal adrenal, kidney and liver. *MDR1* mRNA was not detected by Northern hybridization of breast cancers demonstrating *MDR1* mRNA by RT-PCR although positive control cell lines did demonstrate detectable *MDR1* mRNA. No correlation was found between *MDR1* gene expression by RT-PCR and either with responsiveness to Paclitaxel therapy for which 29 patients were evaluable ($P=0.16$, Fisher Exact Test) or overall survival of 32 breast cancer patients with clinical follow-up information ($P=0.46$, Logrank). The preliminary findings indicate that P-glycoprotein is expressed in breast tumors at low levels predominantly in reactive stromal cells with little if any expression in the carcinoma cells.

Other Genes. We decided that there was a need to explore other mechanisms of drug resistance in order to evaluate the manner in which cancer cells become resistant to drugs. Because of our access to the tissue resource provided by the clinical trials funded by this grant we were able to begin pilot studies of other genes which might be correlated with cytotoxic drug resistance in breast cancer. Two of the genes initially investigated were the p53 tumor suppressor gene and a small molecular weight protein (p7) which is overexpressed in human drug-resistant breast and ovarian cancer cell lines.

P7 was initially discovered in ovarian carcinoma cell lines following exposure to vinblastine or adriamycin treatment *in vitro* (3-6). A similar cell line expressing high levels of p7 was obtained with MCF-7 human breast cancer cells following treatment with doxorubicin and vinblastine chemotherapeutics. The expression of p7 was preliminarily investigated in three groups of women with breast cancer: 1.) frozen primary breast cancers from the USC Breast Tumor and Tissue Bank and obtained from women with known clinical follow-up histories (25 patients), 2.) women entered in a clinical trial of 5-fluorouracil and radiation therapy for locally advanced breast cancer (protocol no. 1B-

93-3) (38 patients) and 3.) breast cancers from women prior to entry in a clinical trial of paclitaxel treatment for advanced breast cancer (protocol no. 1B-92-3) (48 patients).

P7 expression in breast cancer specimens. Among primary, untreated breast cancers p7 was expressed in six of 25 carcinomas (24%) by immunohistochemistry with 1D7 monoclonal antibody. P7 was localized to the cell membrane and cytoplasm of tumor cells. No immunostaining was found in normal breast ductal or lobular epithelium, stromal cells, endothelial cells of blood vessels, lymphocytes or macrophages. In this small cohort of cases p7 expression was not related to histologic grade, tumor size, lymph node involvement, presence of distant metastases at diagnosis, estrogen receptor status, HER-2/neu oncogene expression or P53 tumor suppressor protein expression. p7 was correlated with stage, however this did not reach statistical significance ($p = 0.08$) probably because of the small number of cases in this preliminary study. p7 was correlated with expression of progesterone receptor ($p = 0.035$) and recurrent disease during follow-up ($p = 0.003$) (see Table). These preliminary results appeared to merit additional studies.

Is P7 a prognostic marker and/or as a predictor of responsiveness to conventional therapies in breast cancer? P7 expression was evaluated in 38 women who accrued to an institutional multimodality protocol of continuous infusion 5-fluorouracil (FU) and concomitant radiation therapy for potentially resectable, locally advanced breast cancer. In this experimental protocol, biopsy of breast carcinomas was performed prior to neoadjuvant therapy and resection of the mass was performed after therapy. Neoadjuvant treatment involved pre-operative 5-FU ($200\text{mg}/\text{m}^2$) administration followed by a second incisional biopsy of the remaining breast tumor and then radiotherapy (50 Gy) to the breast and regional lymph nodes followed, finally, by resection of the remaining breast tumor mass. In these women P7 expression was increased following 5-FU treatment and radiation therapy. Overall, expression of P7 protein was increased from 21% of cases showing P7 expression to 44% after 5-FU and radiation therapy treatments. Only one breast cancer which had expression of P7 in less than 5% of tumor cells showed a loss of expression following treatment. The change in expression approached statistical significance ($p = 0.06$) even though the number of cases evaluated was small.

Table 1. Association of p7 Expression with Other Prognostic Factors.

Patient group	No. of patients	No of P7 positive case (%)	P value ¹
Total	25	6 (24)	
Age			
<50 yr	10	3 (30)	0.566
≥50 yr	15	3 (20)	
Tumor size			
<3 cm	6	1 (17)	1
≥3 cm	19	5 (26)	
Histologic grade			
1-2	8	2 (25)	1
3	17	4 (24)	
Stage			
T1/T2	15	1 (7)	0.08
T3/T4	7	3 (43)	
Unknown	3		
Regional node involvement			
positive	16	5 (31)	0.62
negative	8	1 (13)	
Unknown	1		
Distant metastasis			
positive	16	5 (31)	0.13
negative	8	1 (13)	
Unknown	1		
ER status			
positive	9	3 (33)	0.27
negative	12	1 (8)	
Unknown	4		
PR status			
positive	10	4 (40)	0.035
negative	11	0 (0)	
Unknown	4		
P53			
positive	5	2 (40)	0.54
negative	12	2 (17)	
Unknown	8		
HER-2/neu			
positive	8	1 (13)	0.59
negative	10	3 (30)	
Unknown	7		
Clinical outcome²			
AWD	11	6 (55)	0.003
NED	14	0 (0)	

¹Fisher's Exact Test (2-Tail).²AWD: Alive with disease; NED: No evidence of disease.

p7 expression was also characterized in our study of tumor markers as predictors of responsiveness to taxol chemotherapy, clinical trial (1B-92-3). We found a significant correlation between p7 expression in these breast cancer and both overall survival and responsiveness to treatment. After a median follow-up of 7 months, 9 of 9 patients (100%) whose breast cancers expressed p7 died from tumor progression, compared with 23 of 40 patients (55.7%) whose breast cancer did not express p7 ($p = 0.017$).

Among 37 patients, evaluable for response, there were four complete responders (CRs) (10.9%) and 11 partial responders (PRs) (32.4%), for an overall response rate of 43.3%. p53 expression was also investigated but in these cases but only P7 protein was significantly correlated with the response of advanced breast cancer patients to paclitaxel therapy ($P=0.032$). Fourteen of the 15 patients (93%) lacking P7 expression were responders to paclitaxol treatment whereas only 9 of 22 (40.9%) with P7 expression responded.

Our preliminary studies in different cohorts of breast cancer patients, suggested that P7 protein might have clinical utility for predicting tumor progression and response to neoadjuvant therapy.

Clinical Studies.

Protocol #1B-93-8. This clinical study, previously approved by U.S. Army Medical Research and Materiel Command's Human Use Office, was closed for accrual in January 1997. The protocol accrued eight patients into the study, with the last patient being accrued in February 1996, and going off study in May 1996. Table 1 provides the outcome of these patients and comments on their course. In common to many other investigators, the major problems encountered in treating patients in this protocol, were practical issues in relation to continuos infusion paclitaxel exceeding 24 hours. Patients 1 and 4 manifested several episodes of catheter sepsis, and patient 2 went off study because of catheter fracture, and eventually had a chest wall abscess in relation to a retained catheter fragment. This experience led us to explore and develop a protocol to test resistance-reversal strategies employing short-infusion paclitaxel, since a randomized trial employing infusions was not likely to be feasible.

Table 1. Patient demographics and outcome (protocol #1B-93-8)

Patient Number	On Study (months)	Response/Evaluability	Comments
1	5	Stable soft tissue	catheter sepsis
2	16	Stable bone/imp marker	catheter fracture
3	2	Refused therapy/NE	
4	5	partial response/lung	catheter sepsis
5	2	Refused therapy/NE	
6	6	Partial response/skin	
7	1	skin progression/NE	started other therapy
8	3	stable skin	started other therapy

NE=not evaluable

The slow accrual in this study was partly related to changes in the way paclitaxel was usually administered from infusions to short (1 to 3 hours infusions), and partly due to the reluctance in placing central venous catheters in patients at the Los Angeles County Hospital when the episodes of catheter sepsis first became manifest. Possible factors in the high incidence of infections may include the insolubility of paclitaxel leading to not infrequent visible particulate matter, and the predisposition to have some neutropenia. No toxicity was clearly attributable to the high doses of magestrol acetate given by suspension over 6 days. Patient #2, in fact, tolerated the intermittent doses on the drug without the usual weight gain that is seen with this drug. Also, no thromboembolic phenomena were evident. It is noteworthy that two objective responses and one long improvement in bone disease and markers were seen among 5 evaluable patients, inpatients who had already paclitaxel and had shown progressive disease on this drug. This might be related to schedule and not necessarily to resistance reversal since Seidman et al. did report responses to paclitaxel by 96 h infusions, following failure of shorter infusions. However, in ovarian cancer long infusions were ineffective after failure of the shorter schedules (Markman M, personal communications). After the last two patients opted to terminate treatment early, it was decide to close the protocol as it would never lead to a practical, randomized clinical study.

Protocol #1B-95-4. When it became evident that the long term infusion protocol was impractical, we initiated efforts to select a resistance reversal agent that could be given with a short infusion of paclitaxel. Also, we considered practical issues of great importance if one were to be able to mount a randomized study of paclitaxel with and without a resistance reversal agent. We selected PSC-833 as the most promising agent to reverse MDR1, and were successful in obtaining the drug from Sandoz, now Novartis. The protocol underwent several drafts in order to comply with their requirements and seek collaborations with the City of Hope, UC Davis, and Bayside Hospital in Toronto. Except for the City of Hope, others remain to work out their protocols. In the meantime, the rationale for the study changed somewhat from aiming to reverse drug resistance, to perhaps inhibit the emergence of MDR. This new rationale is based on the findings by the laboratory of Branimir Sikic (Beketic-Oreskovic, et al. JNCI 1995; 87:1593-1602) that PSC-833 can inhibit the emergence of MDR in cell lines exposed to doxurubicin, with mutants eventually emerging but the decreased topoisomerase II-alpha rather than MDR1.

To date seventeen patients have been accrued to this study as summarized in the attached table. Ten subjects have entered arm 2 (Paclitaxel + PSC833) and 7 subjects on arm 1 (Paclitaxel alone). Preliminary evaluation of response indicates 4 partial responses, 2 in each arm. The median time on study is 133 days, and 5 subjects continue on study.

No.	On-study Date	Off-study Date	Study Site	On-study Days	Arm	Response	Comments
1	8/19/96		COH	414	2		
2	12/10/96	02/12/97	COH	64	1		
3	8/13/97		COH	55	2		
4	8/18/97		COH	50	2		
5	8/22/96	06/20/97	USC	302	1	PR	
6	10/9/96	10/11/96	USC	2	1		Inevaluable
7	10/29/96	09/04/97	USC	310	1		
8	11/12/96	12/23/96	USC	41	2		
9	12/10/96	02/25/97	USC	77	1		
10	12/16/96	03/17/97	USC	91	2		
11	1/3/97	08/11/97	USC	220	1		
12	1/17/97	04/17/97	USC	90	2		
13	1/21/97	07/02/97	USC	162	2	PR	
14	3/3/97		USC	218	2	PR	
15	3/10/97	06/03/97	USC	85	1	PR	
16	4/30/97	05/21/97	USC	21	2		Inevaluable
17	8/12/97		USC	56	2		

The study, recently submitted to the U.S. Army Medical Research and Materiel Command's Human Use Office, has gone through additional revisions to increased accrual since there is considerable competition for patients. The current format is one of a randomized phase II study, and the major objective is to verify response rates. If feasible, biopsy material will seek to identify baseline tumor characteristics with respect to MDR1, and also whether treatment with paclitaxel leads to MDR1 overexpression. The last question is also being addressed in specimens obtained from other trials using paclitaxel.

Other related clinical studies. Investigators at USC (Dr. Silvia Formenti) and New York University (NYU, Drs. Franco Muggia and Matthew Volm) are collaborating in a study of neoadjuvant paclitaxel for locally advanced breast cancer, under grant support from the California Breast Cancer research Program. To date, NYU has recruited 11 patients into this study and has stored specimens, being batched for subsequent determinations in the laboratory of Dr. Michael Press. Some specimens were sent earlier, but technical difficulties prompted storage of the specimens until they can be safely sent. Among the specimens, we have post treatment samples that will allow us to answer the question

about the presence of P-glycoprotein after treatment with paclitaxel. Also, though collaboration with Dr. Sikic's laboratory we plan to assess the presence of beta-tubulin isoforms that are considered to predict for paclitaxel sensitivity.

Finally, at NYU, work with the Nuclear Medicine Department (Dr. Elissa Kramer) has permitted the measurement of 99Tecnetium sestamibi clearances. Fast clearances have been related to the presence of P-glycoprotein, but many also reflect the function of other ATP binding cassette proteins, such as MRP. The study of such clearance is associated with assessment of P-glycoprotein, MRP, and other markers of resistance may prove of interest, particularly since earlier studies have claimed a correlation of altered clearances with response to treatment with anthracyclines in breast cancer.

CONCLUSIONS.

Taxol chemotherapy was effective in prolonging the survival of some patients in the clinical trial, however, neither P-glycoprotein nor p53 expression in the breast cancer specimens was significantly correlated with responsiveness to drug treatment. A recently described small molecular weight protein, p7, was correlated with responsiveness to treatment and longer overall survival suggesting that this protein merits further investigation in our breast cancer clinical trials.

REFERENCES:

1. Muggia FM. Managing breast cancer in an outpatients setting. *Breast Cancer Res & Treat* 21: 27-34, 1992.
2. Harris J, Veronesi U, Henderson IC. Breast cancer. *New Eng J Med*, 1993.
3. Yang X, and Page M. An Mr. 7 KDa membrane protein overexpressed in human multidrug-resistant ovarian cancer cells. *Cancer Lett.* 88: 171-178, 1995.
4. Yang X, and Page M. Growth regulation of multidrug resistant ovarian cancer cells by 1D7 monoclonal antibody. *Anticancer Res.* 16: 273-276, 1996.
5. Yang X, and Page M. A Mr. 7 kDa protein is present in both plasma and nuclear membranes in multidrug-resistant ovarian cancer cells. *Oncol Rep.* 3: 181-182, 1996.
6. Yang X, and Press M. Unpublished observations.